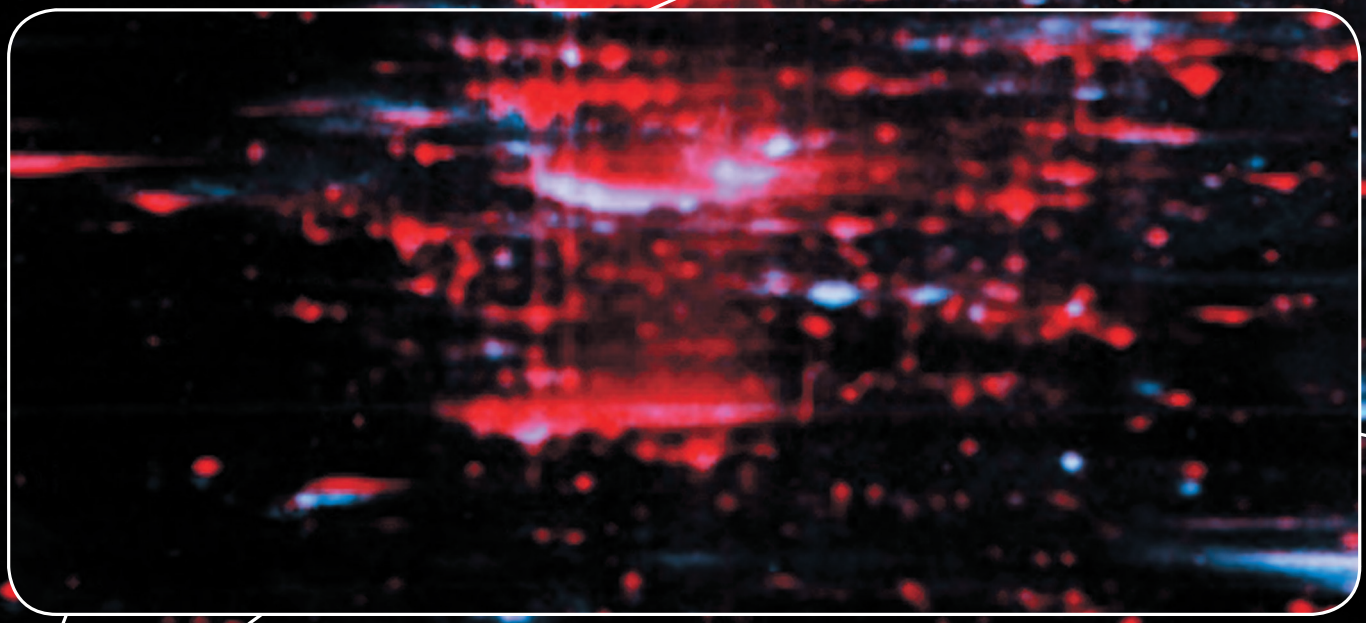
Pro-Q Diamond Phosphoprotein Gel Stain

In-gel Detection Technology for Protein Phosphorylation



IN-GEL DETECTION	Detect phosphoproteins directly in 1-D and 2-D gels
SIMPLE	Just fix, stain and destain — no blotting or antibodies required
VERSATILE	Detect phosphate groups attached to tyrosine, serine or threonine residues
QUANTITATIVE	Signal is linear over three orders of magnitude and correlates with the number of phosphates
COMPATIBLE	Stained proteins can be accurately identified by mass spectrometry
EASY TO VISUALIZE	Suited for many scanning instruments
MULTIPLEXING CAPABILITY	Use with SYPRO Ruby protein stain and Pro-Q Emerald glycoprotein stain for Multiplexed Proteomics analysis

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Technical Information

Ideal for kinase studies and phosphoproteomics, the Pro-Q Diamond phosphoprotein gel stain is a breakthrough technology that provides a simple, direct method for specifically staining phosphoproteins in polyacrylamide gels.[1] This stain can be used with standard SDS polyacrylamide gels or with 2-D gels — there is no need for phosphoprotein-specific antibodies or Western blot detection reagents, and blotting is not required. The stain is also compatible with mass spectrometry, allowing, for the first time, meaningful analysis of the phosphorylation state of entire proteomes.

The stain allows detection of phosphate groups attached to tyrosine, serine or threonine residues and can be visualized using a variety of scanning instruments. The sensitivity limit ranges from ~1–16 ng/band depending on the phosphorylation state of the protein. For individual phosphoproteins, the Pro-Q Diamond signal is linear over three orders of magnitude and the strength of the signal correlates with the number of phosphate groups in the band or spot.

Pro-Q Diamond phosphoprotein gel stain is even more powerful when used together with SYPRO Ruby protein gel stain (S-12000, S-12001, S-21900), a total-protein stain, for multiplexed staining. Because both stains are quantitative, the ratio of the Pro-Q Diamond signal to the SYPRO Ruby signal intensity provides a measure of the relative phosphorylation level of the protein in each band or spot. Pro-Q Diamond phosphoprotein gel stain is also compatible with our Pro-Q Emerald glycoprotein gel stain, for expanded Multiplexed Proteomics analysis.

Materials Supplied

Pro-Q Diamond phosphoprotein gel stain is supplied as a ready-to-use solution in several sizes. A minigel requires ~50 mL and a large-format 2-D gel requires ~500 mL of stain.

Ordering Information

P-33301	Pro-Q Diamond phosphoprotein gel stain	200 mL
P-33300	Pro-Q Diamond phosphoprotein gel stain	1 L
P-33302	Pro-Q Diamond phosphoprotein gel stain	5 L
P-33310	Pro-Q Diamond phosphoprotein gel destaining solution	1 L
P-33311	Pro-Q Diamond phosphoprotein gel destaining solution	5 L

For quantitative multiplexed staining, buy both stains together and save 10%!

P-33305	Pro-Q Diamond phosphoprotein gel stain and SYPRO Ruby protein gel stain pack (1 L each)
P-33306	Pro-Q Diamond phosphoprotein gel stain and SYPRO Ruby protein gel stain pack (200 mL each)

References

1. Steinberg et al., manuscript submitted.

For further information contact

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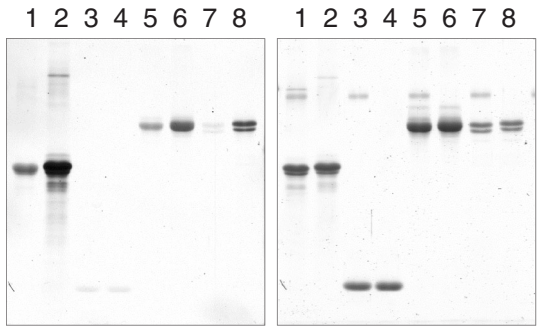
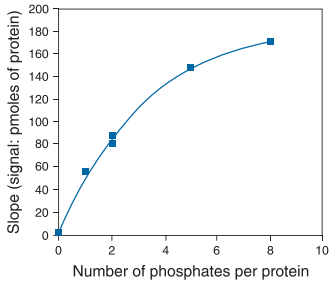
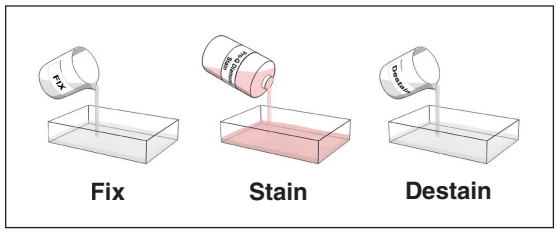


Specificity of Pro-Q Diamond phosphoprotein gel stain. A polyacrylamide gel containing various proteins was stained with Pro-Q Diamond phosphoprotein stain (left) followed by SYPRO Ruby protein gel stain (right). The gel shows the nonphosphorylated protein lysozyme (lanes 3 and 4) and the phosphoproteins α-casein (lanes 1 and 2), ovalbumin (lanes 5 and 6) and pepsin (lanes 7 and 8) before (even lanes) and after (odd lanes) treatment with phosphatases. Pro-Q Diamond staining indicates loss of phosphates (ovalbumin and pepsin), partial loss of phosphates (α-casein) or no change (lysozyme).



Sensitivity and quantitation capabilities of Pro-Q Diamond phosphoprotein gel stain. Dilutions of six different proteins were separated by SDS-polyacrylamide gel electrophoresis and stained with Pro-Q Diamond phosphoprotein gel stain. The image was documented on a Fuji FLA 3000 scanner (Fuji) and the fluorescence emission from each band was quantitated. The fluorescence signal was plotted against the moles of protein and showed linearity over three orders of magnitude for each protein (data not shown). The slope of the line for each protein was then plotted against the known number of phosphates per protein, showing a strong correlation between the two parameters.



Staining with Pro-Q Diamond dye is simple — just fix, stain and destain.

Cover image:
Visualization of total proteins and phosphoproteins in a 2-D gel. Proteins from a Jurkat T-cell lymphoma line cell lysate were separated by 2-D gel electrophoresis and stained with Pro-Q Diamond phosphoprotein gel stain (blue) followed by SYPRO Ruby protein gel stain (red). After each dye staining, the gel was imaged on an FLA-3000 scanner (Fuji). The digital images were overlaid using Z3 software (Compugen) and the resulting composite image was digitally pseudocolored using Adobe Photoshop (Adobe).